Preliminary Studies on the Toxicity and Metabolism of Palladium and Platinum

by W. Moore,* D. Hysell,* L. Hall,* K. Campbell,* and J. Stara*

Preliminary data are given on the LD_{50} of $PdCl_2$ following different routes of exposure and on the LD_{50} of $PtCl_4$ following intravenous exposure.

The retention, tissue distribution, and excretion of [103]Pd and [191]Pt in rats was determined following oral, intravenous, intratracheal, and inhalation exposure. The highest retention for both [103]Pd and [191]Pt was obtained following intravenous dosing, and the lowest retention occurred after oral dosing. Following a single oral dose, almost all of the [103]Pd and [191]Pt was excreted in the feces due to nonabsorption, whereas after intravenous dosing, similar quantities were excreted in both the urine and feces. Tissues containing the highest concentrations of these metals were the kidney, spleen and liver. Following intravenous dosing of pregnant rats, a small amount of [109]Pd and [191]Pt was found in the fetuses.

Introduction

Automotive manufacturers have indicated that palladium (Pd) will be used in connection with platinum (Pt) in automotive catalytic converters. These converters are designed to reduce the concentrations of carbon monoxide (CO) and hydrocarbons (HC) in the exhaust stream by oxidizing them into carbon dioxide and water. The control of the concentrations of CO and HC in automotive emissions is necessary for light-duty vehicles to comply with the CO and HC emission standards set forth in the Clean Air Amendments of 1970 (1). With the use of Pd and Pt in automotive catalytic converters, there is the possibility that some of the material will be emitted to the atmosphere or

enter into other segments of the environment following degradation or disposal of worn-out converters.

At the present time, the authors are not aware of any information concerning the chemical form of Pd or Pt which may be emitted in the exhaust. It can be speculated that the attrition products from the catalyst could include: particulate composed of the metals combined with substrate material; Pd and Pt oxides; Pd and Pt metals; possibly other chemical forms.

Although toxicological data on these two metals is extremely limited, reports indicate that toxicity varies with the chemical form (2–9). In the studies that are discussed today, we have used the soluble salts of Pd and Pt in order to ascertain some of the basic toxicological and metabolic aspects. It is realized that the availability and metabolism of other chemical forms or substrate material containing Pd and Pt may be different; however, these data should serve as

* U.S. Environmental Protection Agency, National Environmental Research Center, Environmental Toxicology Research Laboratory, Cincinnati, Ohio 45268.

a guideline for additional studies on the biological effects of these two elements.

Methods

The outbred albino rats (Charles River CD–1–strain) used in these studies were maintained on a commercial diet (Purina Lab. Chow) and tapwater *ad libitum* except where otherwise noted. Rabbits were obtained from a local supplier and fed a commercial diet (Purina Rabbit Chow) and tapwater *ad libitum*.

Toxicity Studies of Pd and Pt

Animals were given $PdCl_2$ or $PtCl_4$ by the following routes: orally (PO), intravenously (IV), intraperitoneally (IP); and intratracheally (ITR). All solutions were prepared in saline with no pH adjustment.

Four groups (10 animals/group) of rats were given Pd or Pt in the drinking water (pH 3.0), and appearance, body weight, and water consumption were noted. The concentrations used were 92 and 184 ppm K_2PdCl_4 and 235 and 470 ppm K_2PtCl_4. Water for control animals also had a pH 3.0.

Metabolism Studies of Pd and Pt

The various treatment groups consisted of those (rats) who received Pd or Pt by the following routes: ITR, PO, IV, or by inhalation.

Intratracheal Administration: Ten fasted male rats weighing 180–200 g, were anesthetized and the trachea isolated through a ventral midline incision. $^{103}PdCl_2$ (25 μCi in 0.1 ml saline) was injected and the incision closed. Rats were counted for whole body retention of ^{103}Pd.

Oral Administration: Twenty fasted male rats, 180–200 g, were lightly anesthetized with ether and given 25 μCi of $^{103}PdCl_2$ in 0.2 ml saline by stomach tube. Ten rats were placed in metabolism cages for collec-

tion of 24-hr urine and fecal samples to determine routes of excretion and for determination of whole-body retention of ^{103}Pd. The other 10 rats were sacrificed 24 hr following dosage to establish organ distribution of the ^{103}Pd. Fifteen nonfasted suckling rats, 30 g, were given a single dose of $^{103}PdCl_2$ (25 μCi in 0.2 ml saline) by stomach tube. These animals were maintained for comparison with retention of ^{103}Pd in the adult rats.

Intravenous Administration: Twenty male rats, 180–200 g, were given 25 μCi $PdCl_2$ in 0.1 ml saline IV in a tail vein. Ten rats were sacrificed 24 hr later for organ distribution; 10 rats were placed in metabolism cages for collection of 24-hr samples of urine and feces and for whole body counting to determine retention. Thirteen female rats (16 days pregnant) were given 25 μCi $^{103}PdCl_2$ IV and maintained in metabolism cages for collection of feces and urine. They were sacrificed 24 hr after dosage for determination of organ distribution and placental transfer of the ^{103}Pd.

Inhalation Administration: The animal exposure system consisted of a multiple-animal exposure chamber (88 rats) supplied with aqueous aerosol from a nebulizer; the aerosol droplets passed through a drying cylinder to produce the dry solid particles needed for the inhalation exposure.

The exposure to a platinum particulate involved a 5.8 ml supply of the nuclide $^{191,193}Pt$, assaying 7 mCi in the form of $^{191}Pt^{+4}$ in $1M$ HCl; it was mixed with stable $PtCl_4$ in aqueous solution to give a final 10 ml volume of approximately 2.2 mg/ml. On the basis of such a concentration value and of a median droplet diameter of 6–8 μm generated by the nebulizer, it was calculated that a dry particle of $PtCl_4$ with an aerodynamic size diameter of nearly 1.0 μm would be obtained. The particulate load was measured at 5.0 mg/m^3 over the 48-min exposure period.

The exposure to palladium particulate made use of a 4 ml supply of the nuclide ^{103}Pd, assaying 5 mCi in the $PdCl_2$ form in $0.6M$

HCl. The radioactive form was mixed with stable PdCl₂ in a similarly HCl-acidified solution to give a final 10 ml volume of approximately 2.3 mg/ml. A 1.0 μm aerodynamic diameter was intended. Since the radioactive-stable mix solution was aerosolized in just 30 min, the atmospheric concentration produced was measured as 7.2 mg/m³.

Sacrifice and Tissue Sampling

All rats were euthanatized with an overdose of chloroform anesthesia. Samples collected routinely were blood, heart, lung, liver, kidney, adrenal gland, pancreas, abdominal fat, spleen, skeletal muscle, bone, brain, testicle, and ovary. In the pregnant females, four placentas, and four fetuses were taken from each pregnant rat, and a pooled sample of fetal livers was also saved.

Radioactive Determinations

[103]Pd in 0.6M HCl was diluted as indicated and used in all the studies. The isotope has a half-life of 17 days and a 0.362 MeV γ. Immediately after dosing, whole-body counts were made on all animals used in the retention studies. The animals were counted daily for the first few days, and then every other day for the duration of the experiment. A 200-channel γ spectrometer with a 5-in. NaI (Tl) crystal was used for whole-body counts. Tissue, urine, and feces samples were counted in a well-type, refrigerated scintillation spectrometer.

[191,193]Pt in 0.5M HCl was diluted and used as described in the Pd studies.

The solution contained at least 50% [191]Pt, and only the [191]Pt γ was counted. All values were corrected for decay ([191]Pt has a half-life of 3 days).

Results

Palladium Toxicity

By using the method of Deichman and LeBlanc (10), the approximate LD₅₀ of PdCl₂

Table 1. Acute lethal toxicity of PdCl₂.

Species	Approx. LD₅₀, mg/kg	Route
Rat	5	IV
Rat	70	IP
Rat	200	PO
Rat	6	ITR
Rabbit	5	IV

was determined for IV, IP, ITR, and PO routes of administration. The results are shown in Table 1. Marked differences were noted among the different routes of administration, ranging from 5 mg/kg for IV to greater than 200 mg/kg for PO.

Using the more precise method of Litchfield and Wilcoxon (11), the IV and IP LD₅₀ (14 days) were determined. The LD₅₀ (14 days) was calculated to be 3.0 mg PdCl₂/kg with 95% confidence limits of 2.57–3.49. The slope was found to be 1.43 with a 95% confidence limit of 1.15–1.77. The χ^2 test indicated that the data were not significantly heterogenous. Following IP administration, the LD₅₀ was calculated to be 123.0 (91.1–166.1) mg PdCl₂/kg with a slope of 1.84 (1.04–3.27); no significant heterogenicity was noted.

Following acutely toxic IV doses of PdCl₂, death occurred very rapidly, with a sharp threshold such that if exitus did not occur within 5–10 min, the animals (both rats and rabbits) survived the 14-day experimental period. Clonic and tonic convulsions were noted in rats and rabbits. Following IP injection, necropsy findings indicated a chemical type "burn" of the viscera in animals dying within 24 hr. Gross pathologic examination of IP dosed survivors at 14 days showed prominent peritonitis with numerous visceral adhesions.

A limited number of rats from the intravenous and intraperitoneal studies were housed in metabolism cages and several toxicometric parameters were measured during the 14-day observation period. Survivors of an acutely toxic IV dose of PdCl₂ exhibited a 25% decrease in water intake and urine

Compound	LD$_{50}$, mg/kg (95% confidence)	Slope (95% confidence)	LD$_{50}$, μM/kg
PdCl$_2$	3.0 (2.6-3.50)	1.43 (1.1 -1.8)	16.9
K$_2$PdCl$_4$	6.4 (6.0-6.8)	1.14 (0.83-1.2)	19.6
(NH$_4$)$_2$PdCl$_4$	5.6 (4.9-6.4)	1.31 (0.96-1.8)	19.7

excretion. Following intraperitoneal dosing, a 7% reduction in body weight was observed with up to 80% reduction in food intake. Water intake was markedly reduced initially and then returned to control levels or above. Proteinuria was noted in all animals following both routes of administration. Elevated urinary ketone bodies were observed in some animals following both routes of dosing.

In order to ascertain the effect of chemical form on toxicity, the LD$_{50}$ of K$_2$PdCl$_4$ and (NH$_3$)$_2$PdCl$_4$ were determined (Table 2). The LD$_{50}$ when expressed in micromoles of Pd was very similar for the three chemical forms.

In the two groups of rats maintained for 33 days on drinking water containing 92 ppm and 194 ppm K$_2$PdCl$_4$, respectively, there were no abnormalities noted in general appearance, body weights or urinalysis.

Platinum Toxicity

The results of a preliminary range finding study on the acute toxicity of IV PtCl$_4$ in rats is given in Table 3. The high incidence of mortality at the lowest dose precluded determination of the LD$_{50}$ (14 days).

However, the lowest dose would appear to be a reasonable approximation.

In two groups of rats maintained for 23 days on drinking water containing 235 ppm and 470 ppm K$_2$PtCl$_4$, respectively, there was a decrease in weight gain and water consumption. For the high dose level the weight gain decreased 14.7% and the water intake decreased 32.8%. No gross pathological changes were found at necropsy. Additional studies are currently in progress on Pt toxicity.

Table 3. Acute intravenous toxicity of PtCl$_4$ in rats.

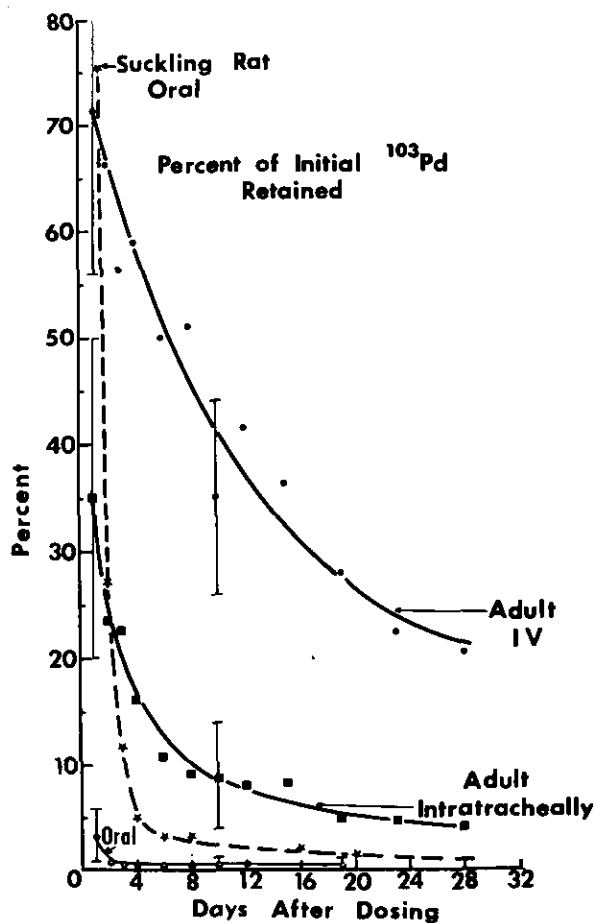| PtCl$_4$ dose, mg/kg	No. of rats per group	Cumulative deaths	Mortality, %
41.4	10	10	100
36.7	10	9	90
31.4	10	9	90
26.2	10	4	40

FIGURE 1. Whole body retention in adult rats of ^{103}Pd following PO, IV, ITR administration and whole body retention in suckling rats following PO administration.

Metabolism of Palladium

Whole-Body Retention. Analysis of the data for whole-body retention of [103]Pd following a single exposure disclosed that the route of administration of the dose significantly affected whole-body retention. The [103]Pd retained in rats, 1 to 28 days after dosing PO, IV, or ITR is presented in Figure 1. Following PO dosing, the retention curve declined very rapidly during the first 3 days to about 0.4% of the initial dose. The initial rapid clearance is attributed to passage of the unabsorbed [103]Pd through the gastrointestinal tract. Extrapolation of the second component of the retention curve to the intercept indicated that absorption was less than 0.5% of the initial dose. Retention of [103]Pd by the suckling rats following oral administration was similar to the adults; however, the amount absorbed and retained with time was significantly higher.

The amount of [103]Pd retained following ITR dosing was significantly higher than for PO dosing and also higher than inhalation exposure (Fig. 2). The difference in retention between inhalation and ITR exposure may be due in part, to protein binding of the [103]Pd following injection. The greatest amount of [103]Pd retained over a period of time occurred following IV administration.

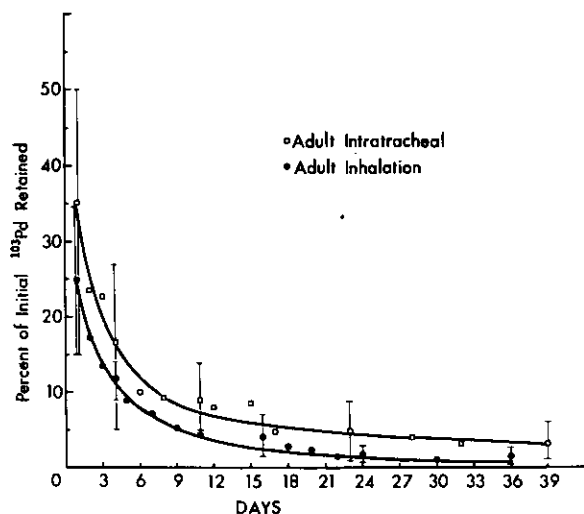
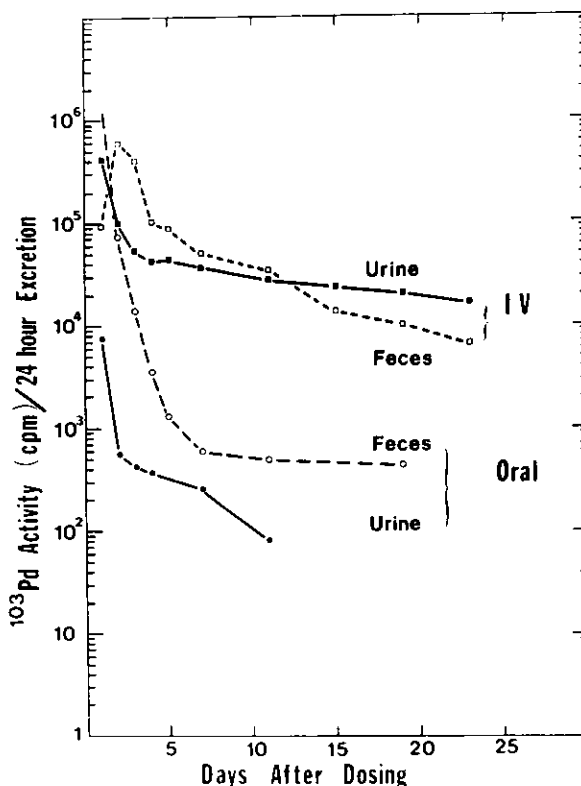FIGURE 2. Whole-body retention of [103]Pd in adult rats following ITR and inhalation exposure.

FIGURE 3. Excretion of [103]Pd by adult rats following PO and IV administration.

Excretion: Radioactive counts of 24-hr urine and feces samples from the rats receiving the [103]Pd PO showed that almost all of the [103]Pd was eliminated in the feces, and only a trace amount was excreted in the urine (Fig. 3). With IV administration, [103]Pd was eliminated both in the urine and feces in similar quantities. Toward the end of the study, urinary excretion exceeded fecal excretion.

Tissue Distribution: The distribution and concentration of [103]Pd was determined for different tissues following PO and IV dosing. Twenty-four hours after PO dosing, detectable quantities of [103]Pd were found only in the kidney and liver. The concentration in the kidney was much greater than in the liver. Twenty-four hours after IV dosing, [103]Pd was found in all the tissues analyzed, with the highest concentrations occurring in

the kidney, spleen, liver, adrenal gland, lung, and bone, respectively.

The rats used in the whole-body retention study were sacrificed 104 days after exposure and the tissues counted. No significant amounts of ^{103}Pd were found in any of the tissues from the group receiving the PO dose. In the IV-dosed rats, the highest concentrations of ^{103}Pd were found in the spleen, kidney, liver, lung, and bone. For the ITR-dosed rats, the lung contained the most ^{103}Pd followed by kidney, spleen, bone, and liver.

Maternal/Fetal Uptake: Thirteen pregnant rats (16th day gestation) were each given 25 μCi ^{103}Pd IV and sacrificed 24 hr later. During the 24-hr period, the pregnant rats excreted 44.2% of the initial IV dose. The amount excreted by the pregnant rats was higher than the amount excreted by the fasted adult male rats during the first 24-hr period. The magnitude of the difference in ^{103}Pd concentration among the maternal organs and also between maternal tissues and the fetuses is best shown by the counts per gram of tissue, which are given in Table 4.

Table 4. ^{103}Pd in maternal organs and fetuses.

Tissue	Mean counts/g
Kidney	588,479
Liver	319,153
Ovary	29,625
Lung	29,21v
Bone	18,351
Blood	3,654
Placenta	58,321
Fetal liver	1,429
Fetus	757

The pattern of distribution and concentration of ^{103}Pd in maternal organs was similar to that previously found in the whole-body IV experiment. Most of the fetuses (35) contained a small amount of ^{103}Pd, and the mean value for these fetuses is given in Table 4. However, radioactive counts for 17 fetuses from five litters was not significantly higher than background. The same pattern of results was obtained for the fetal livers. The amount of ^{103}Pd found in the fetuses
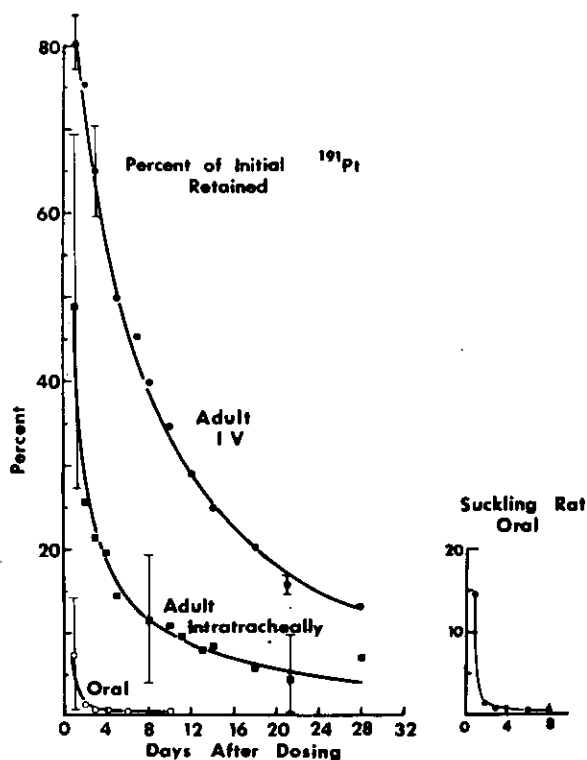

FIGURE 4. Whole body retention of ^{191}Pt in adult rats following PO, IV, ITR administration. Also given is the whole body retention in suckling rats following PO administration.

indicated that Pd does not readily move across the placental barrier in the rat.

Metabolism of Platinum

Whole-Body Retention: The whole-body retention of ^{191}Pt following a single exposure was significantly affected by the route of administration. The per cent of ^{191}Pt retained in rats 1–28 days after dosing PO, ITR, and IV is presented in Figure 4. Following oral dosing, the total net gastrointestinal excretion was extremely high, resulting in a rapid decline of the retention curve to less than 1% at the end of 3 days. The data indicated that the rapid clearance was due to passage of unabsorbed ^{191}Pt through the gastrointestinal tract. Extrapolation of the second component of the retention curve to the intercept indicated that less than 1% of the initial dose was absorbed.
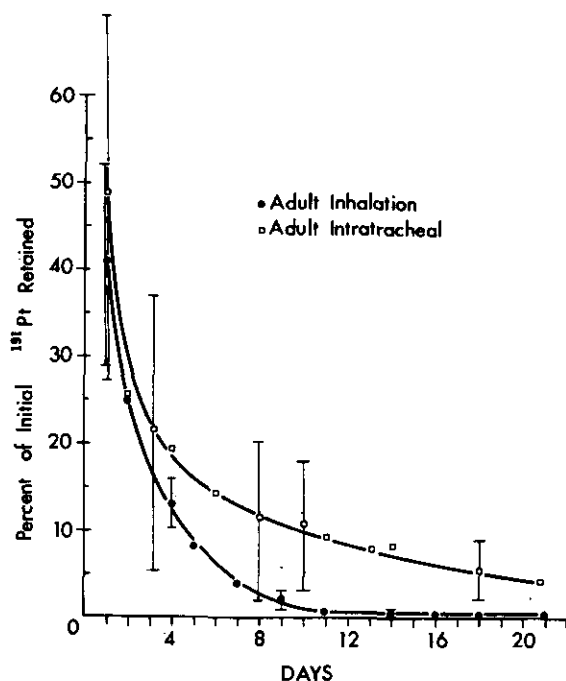
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FIGURE 5. Whole body retention of [191]Pt in adult rats following ITR and inhalation exposure.

FIGURE 6. Excretion of [191]Pt in rats following PO and IV administration.

The whole-body retention of [191]Pt following ITR dosing was significantly higher than for oral dosing. The excretion of approximately 50% of the initial dose during the first 24 hr is attributed to mucociliary and alveolar clearance. The retention of [191]Pt following ITR administration was higher than following inhalation exposure (Fig. 5). Whole-body retention was the highest following IV dosing; the short half-life precluded an accurate determination of the biological half-life.

Excretion: Radioactive counts of 24-hr urine and feces samples from rats receiving [191]Pt PO indicated that almost all of the [191]Pt was eliminated in the feces and only a small amount was excreted in the urine (Fig. 6). These values support the whole-body data that showed that total net gastrointestinal absorption was low. Following IV administration, [191]Pt was excreted in both the urine and feces, but the urine contained a greater quantity. · .

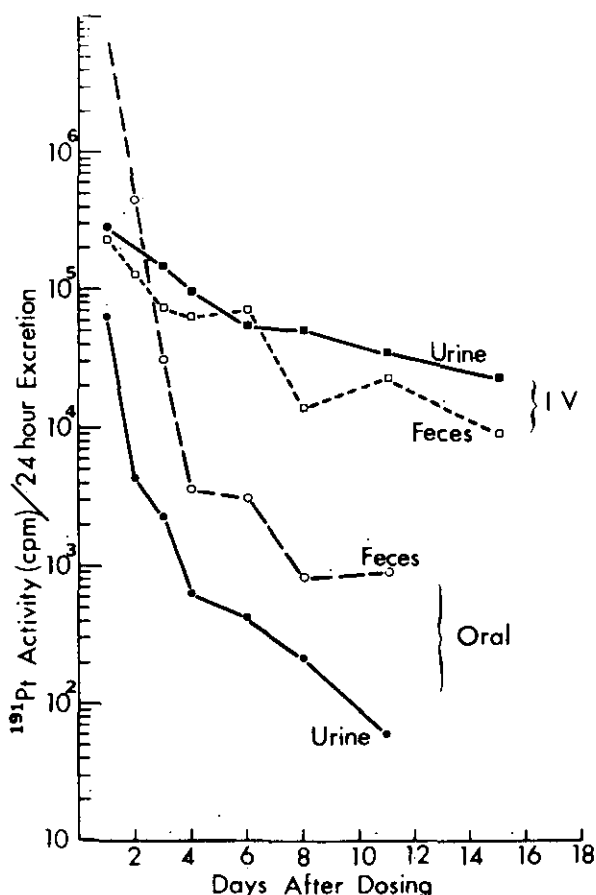*Tissue Distribution:* The distribution and concentration of [191]Pt in tissues was determined following PO and IV dosing. After a single PO dose, the kidney and liver contained the highest concentrations of [191]Pt. The amount of radioactivity found in the other organs was not significantly higher than background. Twenty-four hours after IV dosing, [191]Pt was found in all the tissues analyzed, although most tissues did not contain concentrations of [191]Pt which were appreciably higher than that found in blood. The large amount of [191]Pt found in the kidney suggests that this organ accumulates this element. Concentrations higher than the blood values were also found in the liver, spleen, and adrenal gland. The lower count for the brain suggested that [191]Pt can be

Table 5. ¹⁹¹Pt in maternal organs and fetuses.

Tissue	Mean courts/g
Kidney	127,064
Liver	43,375
Lung	17,981
Ovary	14,639
Blood	10,568
Bone	9,193
Brain	792
Placenta	27,750
Fetal liver	1,421
Fetus	432

transferred through the blood-brain barrier only to a limited extent, possibly the circulating ¹⁹¹Pt is complexed to large molecules that do not cross the blood-brain barrier.

Maternal/Fetal Uptake: Fifteen pregnant rats (18th day gestation) were given 25 μCi ¹⁹¹Pt IV and sacrificed 24 hr later. During the 24-hr period, the pregnant rats excreted 18.8% of the initial dose. The amount excreted by the pregnant rats was approximately the same as the amount (19.3%) excreted by the adult male rats during the first 24-hr period. The concentration of ¹⁹¹Pt per gram for different maternal tissues and fetuses is given in Table 5. The data indicated that there was some transplacental passage of ¹⁹¹Pt; however, there appeared to be placental binding or accumulation. ¹⁹¹Pt was present in all the fetuses (60) counted.

Discussion

It has been stated that the use of catalytic converters will involve exposing the general population to low levels of Pt and Pd compounds and that the exposure to Pt compounds are likely to range from 1/100 to 1/10 of the threshold limit value for Pt salts (12). The threshold limit value (TLV) for inhalation of soluble Pt is 2 μg/m³. Data in the literature suggest that the toxicity of Pt is influenced by the chemical form; the chemical forms of Pt and Pd associated with automotive emissions and the chemical forms that may exist in the environment have not

been adequately identified. The soluble salts of Pd and Pt were selected for initial study because of the paucity of information on the toxicity and metabolism of these ions. In future studies, the biological effects of the metals, oxides of the metals and catalytic attrition products containing Pd and Pt will be investigated.

Although there would be some dermal exposure, inhalation and ingestion would probably comprise the major routes of human exposure to Pt and Pd. The results of these studies indicate that Pt and Pd are not readily absorbed following ingestion. The data also indicate that absorption may be slightly higher in the very young. Following inhalation, somewhat more Pt and Pd were absorbed and retained in the animals. There was some transplacental passage of Pt and Pd but the significance to fetal development is not known. It is obvious from these observations that additional information is needed in order to determine the health impact of these metals in the environment.

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